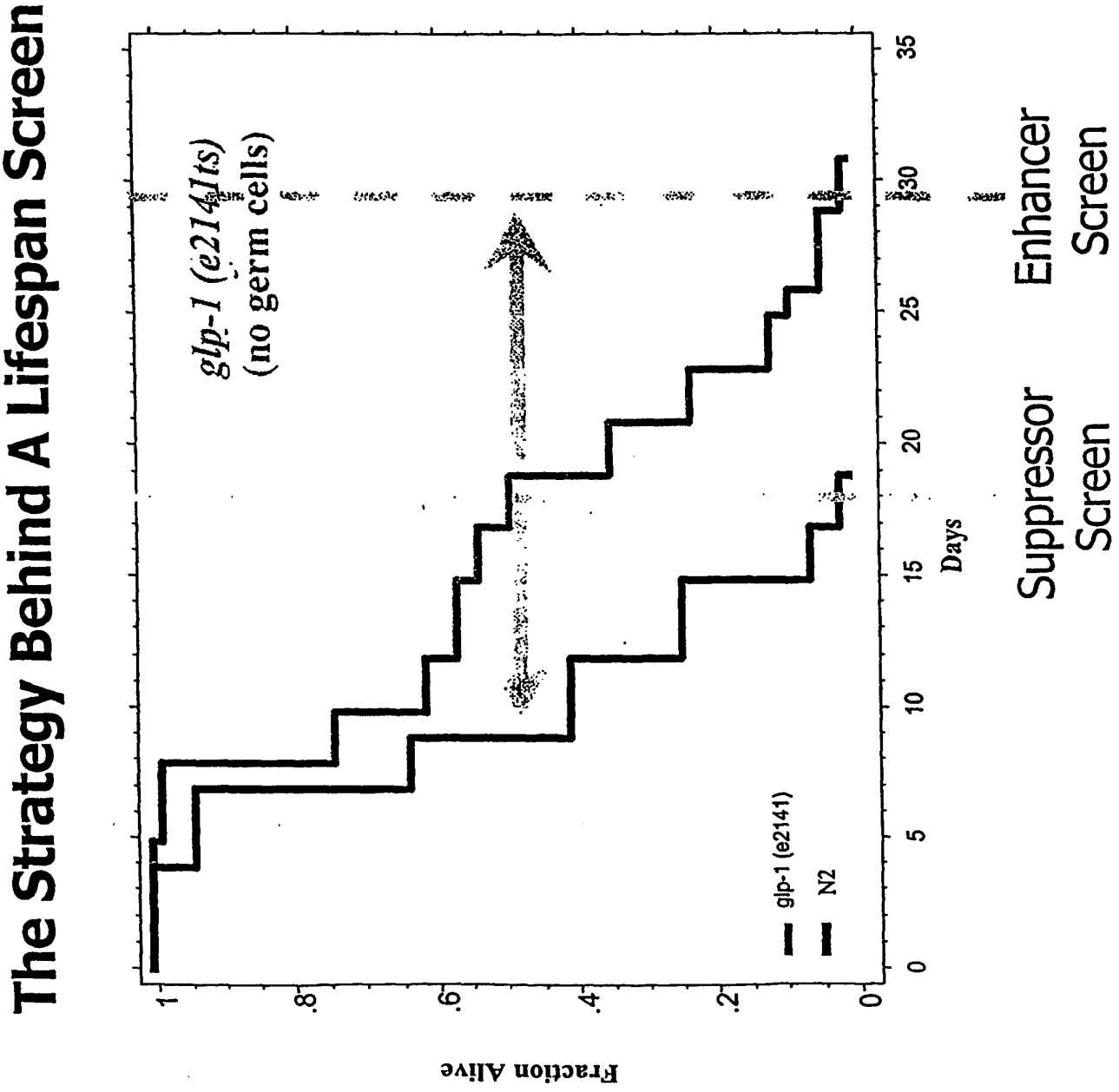
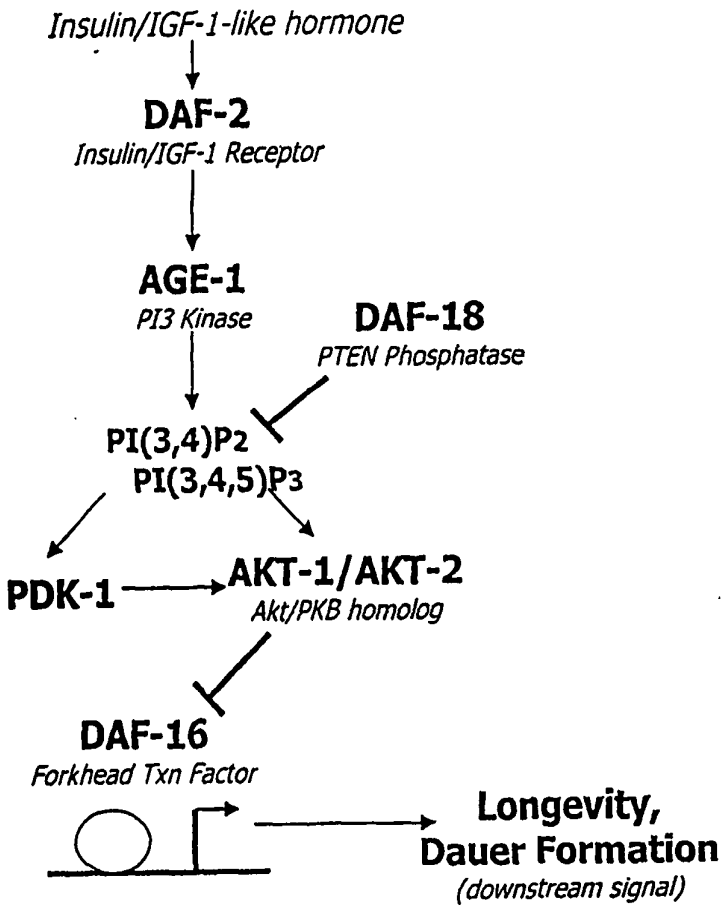

The Strategy Behind A Lifespan Screen
glp-1 (e2141ts)
(no germ cells)
glp-1 (e2141)
N2
Fraction Alive
Days
Suppressor Screen
Enhancer Screen

Figure 1

An Insulin Signaling Pathway and Signals from the Reproductive System Regulate Lifespan in *C. elegans*



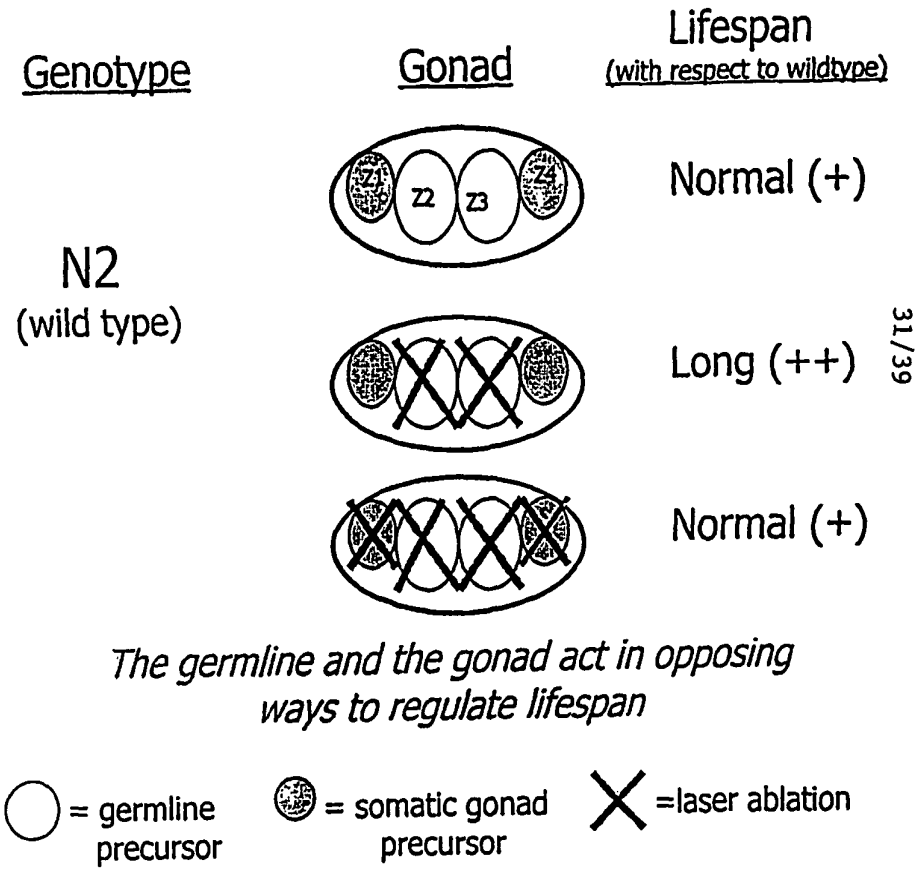
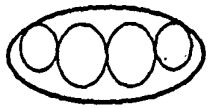
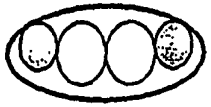


Figure 2

The Reproductive System Modulates Several Genes to Control Aging

Genotype	Gonad	Lifespan (with respect to wildtype)
daf-16(-) or *daf-12(-)* or *daf-9(-)*		Short (-)
		Short (-) *[no change]*

The germline signal shortens lifespan through downregulation of daf-16, daf-12, and daf-9

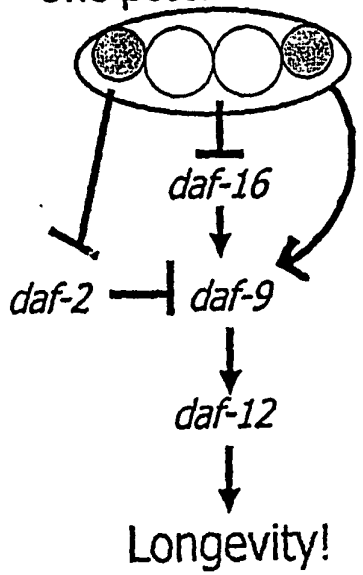
Genotype	Gonad	Lifespan (with respect to wildtype)
daf-2(-) [strong alleles]		Long (++)
		Even Longer (+++)
		Even Longer (+++)

The somatic gonad, but not the germline, extends life through modulation of the daf-2 insulin pathway.

daf-16(-)		Even Shorter (--)
daf-12(-) or *daf-9(-)*		Short (-) *[no change]*

The somatic gonad signal acts through daf-12 and daf-9, yet independently of daf-16, to promote longevity.

One possible model:

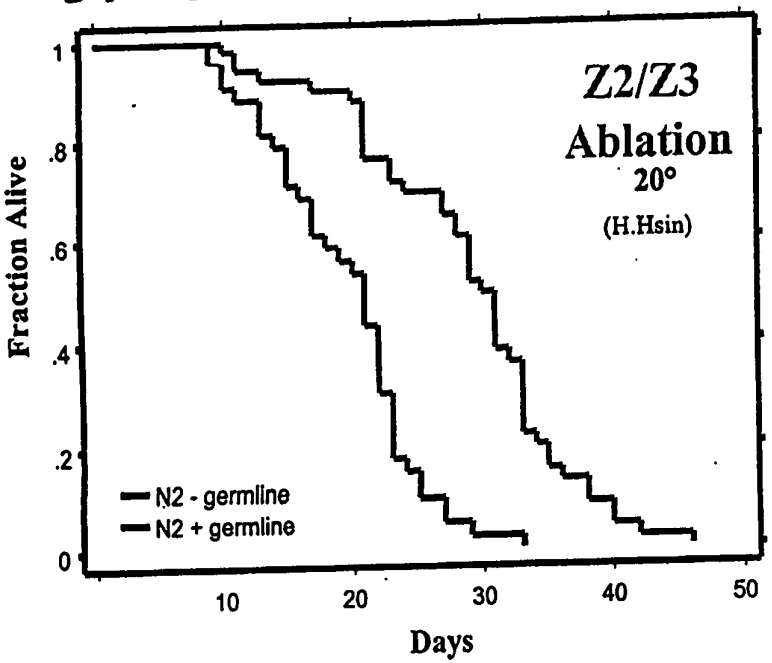


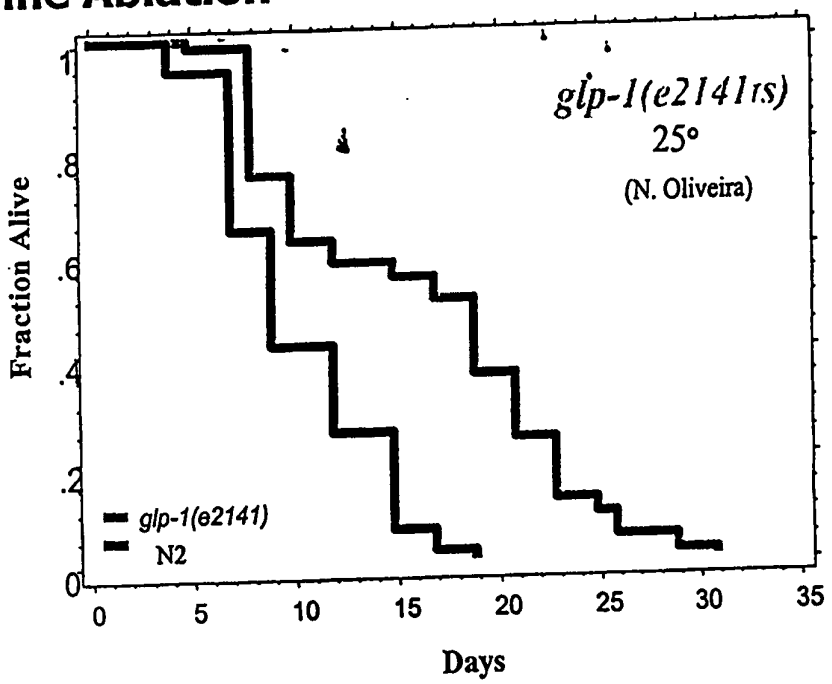
What other genes are involved in this process?

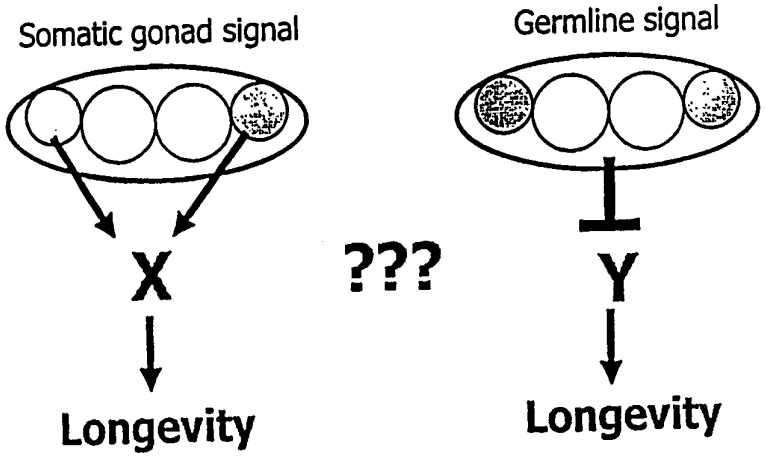
Figure 3

An RNAi Screen for Suppressors and Enhancers of *glp-1* Lifespan Extension

glp-1(e2141ts) Phenocopies Germline Ablation







Why screen?
To identify novel members of the germline, somatic gonad, and other pathways.

Figure 4

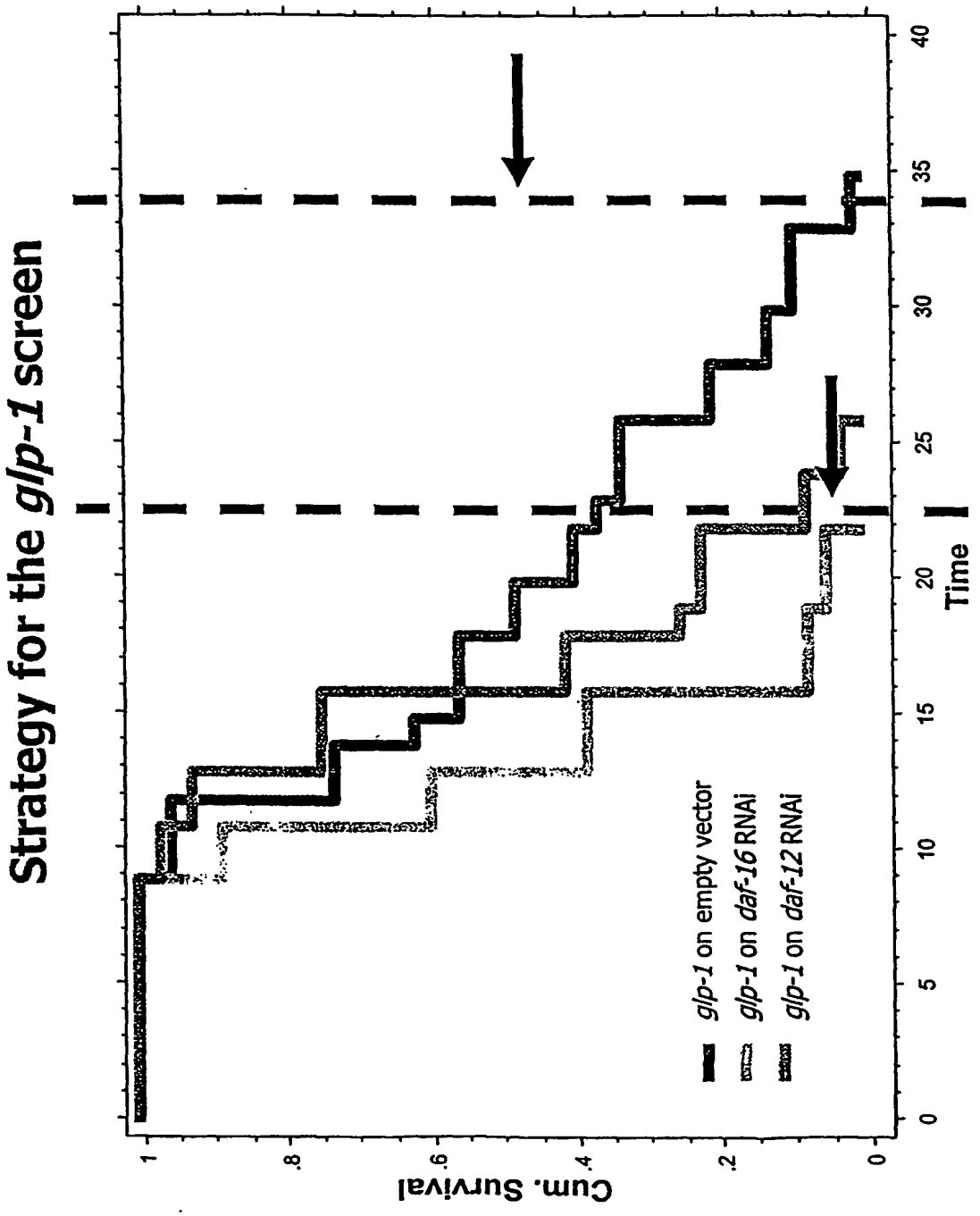
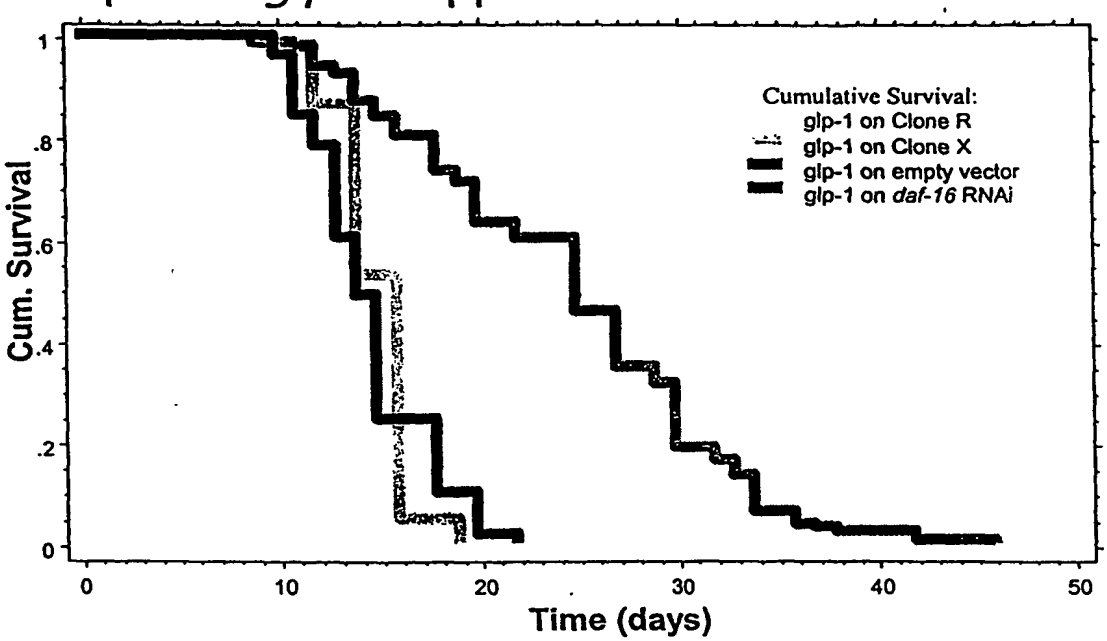

Strategy for the glp-1 screen
Cum. Survival
Time
glp-1 on empty vector
glp-1 on daf-16 RNAi
glp-1 on daf-12 RNAi
~Day 22
Suppressor Screen
~Day 34
Enhancer Screen

Figure 5

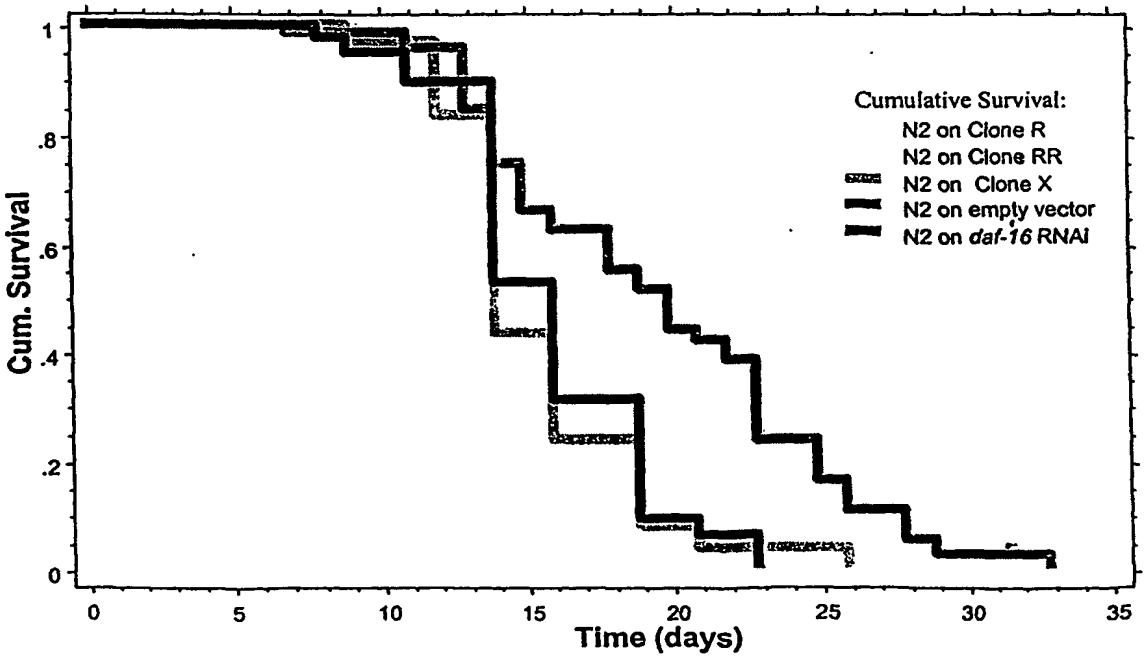
Establishing Specificity: Effects on N2 Longevity

Examples of *glp-1* suppressors:



RNAi Clone R and Clone X suppress *glp-1* longevity to *daf-16* levels

Differential effects on N2 longevity:



Clone R has no effect on N2 lifespan, and Clone X slightly suppresses wild-type longevity to *daf-16* levels. Clone RR dramatically reduces N2 lifespan.

Figure 6

Figure 7

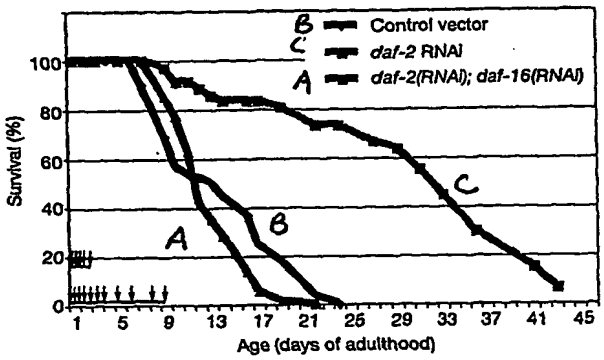

B
C
A
Control vector
daf-2 RNAi
daf-2(RNAi); daf-16(RNAi)
Survival (%)
100
80
60
40
20
0
1
5
9
13
17
21
25
29
33
37
41
45
Age (days of adulthood)
A
B
C

Figure 8

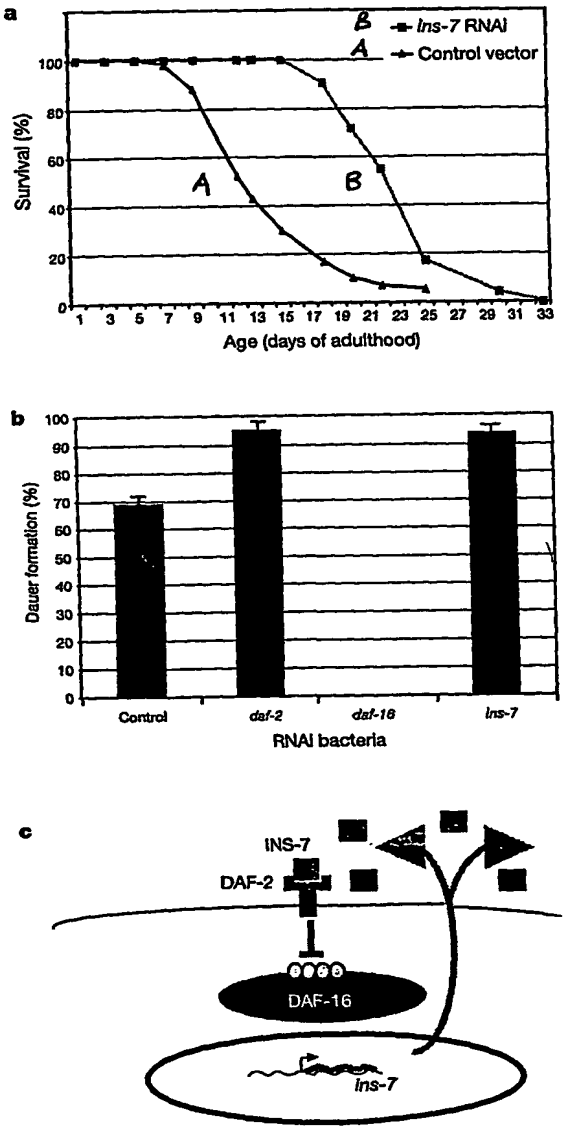

a
B
ins-7 RNAi
A
Control vector
Survival (%)
100
80
60
40
20
0
A
B
1 3 5 7 9 11 13 15 17 19 21 23 25 27 29 31 33
Age (days of adulthood)
b
Dauer formation (%)
100
90
80
70
60
50
40
30
20
10
0
Control
daf-2
daf-16
ins-7
RNAi bacteria
c
INS-7
DAF-2
DAF-16
ins-7

Figure 9

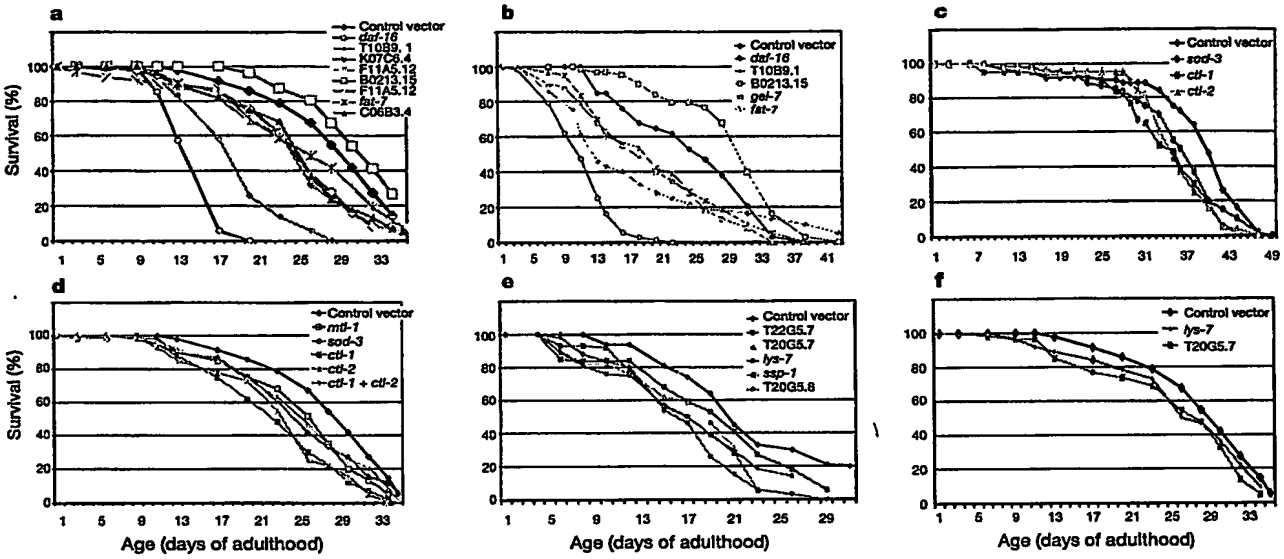
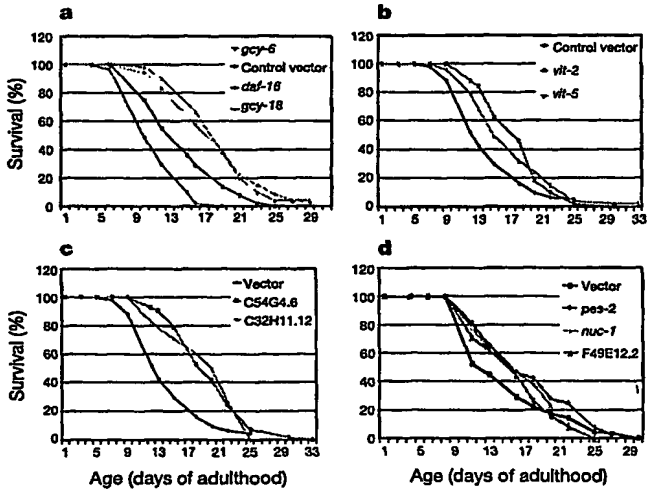

a
Control vector
daf-16
T10B9.1
K07C9.4
F11A5.12
B0213.15
F11A5.12
fat-7
C06B3.4
Survival (%)
b
Control vector
daf-16
T10B9.1
B0213.15
gei-7
fat-7
c
Control vector
sod-3
ctl-1
ctl-2
d
Control vector
mtl-1
sod-3
ctl-1
ctl-2
ctl-1 + ctl-2
Survival (%)
Age (days of adulthood)
e
Control vector
T22G5.7
T20G5.7
lys-7
ssp-1
T20G5.8
Age (days of adulthood)
f
Control vector
lys-7
T20G5.7
Age (days of adulthood)

Figure 10


a
b
c
d
Survival (%)
Age (days of adulthood)
gcy-6
Control vector
daf-16
gcy-18
Control vector
vit-2
vit-5
Vector
C54G4.6
C32H11.12
Vector
pes-2
nuc-1
F49E12.2